84-2 - Salmonella Mutagenicity Test

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DATA EVALUATION REPORT

Study Type: Gene Mutation in Bacteria

TOX. CHEM. No.: 298C

Accession No.: 7E3489

MRID No.:

Test Material: CGA 154281 Technical (93.9% Purity)

Study Number(s): 860840

Sponsor: CIBA-GEIGY Corp.

Test Facility: Experimental Pathology Laboratories, CIBA-GEIGY Limited
Basle, Switzerland

Title of Report:
Samonella/Mammalian-Microsome Mutagenicity Assay (Ames Assay)

Author(s): E. Deparade

Report Issued: September 15, 1986

Conclusions:

CGA 154281 Technical is mutagenic in Ames Test at the concentrations of 2000, 4000, and 8000 ug/plate under the nonactivation assay and at the concentrations of 4000 and 5000 ug/plate under the activation assay.

Concentrations tested: 20, 78, 313, 1250, and 5000 ug/plate (1st and 2nd Trials); 500, 1000, 2000, 4000, and 8000 ug/plate (3rd Trial).

Classification of Data: Acceptable

Title of Report: Salmonella/Mammalian Microsome Mutagenicity Test with CGA 154281 Technical

Procedure:

Five histidine-requiring strains of Salmonella typhimurium (TA98, TA100, TA102, TA1535, and TA1537) obtained originally from Dr. Ames were used in this study.

The mutagenicity of CGA 154281 Technical dissolved in acetone at predetermined concentrations (i.e., 20, 78, 313, 1250, and 5000 ug/plate) was evaluated by the Salmonella/Mammalian Microsome Mutagenicity Test described by Ames (Mutation Res. 31. 347-364, 1975). In the additional experiment carried out with strain TA98 only, the concentrations were 500, 1000, 2000, 4000 and 8000 ug/plate. Mutations were quantified on triplicate plates for each strain by counting His+ revertant colonies after 48 hours of incubation at 37 C. on a minimu medium. Positive controls and solvent control were run concurrently. If the compound was mutagenic, it would demonstrate 2-fold increases over the control value.

Results:

Treatment	Conc. (Per Plate)	Mean Number of His+ Revertant Colonies per Plate									
		TA98		TA100		TA102		TA1535		TA1537	
		+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9
1st Trial:											
Solvent Control		46	31	100	154	242	275	11	15	18	11
CGA 154281	20 ug	42	25	103	151	246	274	16	14	16	9
	78 "	50	30	94	148	217	240	13	13	14	5
	313 "	51	31	109	146	223	226	15	16	19	7
	1250 "	58	35	113	158	179	152	11	15	16	9
	5000 "	56	31	119	117	217	139	16	22	14	12
Positive Controls:											
DNRB-HCl	5 ug	-	355*	-	-	-	-	-	-	-	-
4-NTQLO	0.125 "	-	-	-	596*	-	-	-	-	-	-
MTMC-C	0.5 "	-	-	-	-	-	1031*	-	-	-	-
	1.0 "	-	-	-	-	-	1345*	-	-	-	-
Sodium Azide	2.5 "	-	-	-	-	-	-	-	686*	-	-
	5.0 "	-	-	-	-	-	-	-	988*	-	-
ANAD-HCl	50 "	-	-	-	-	-	-	-	-	-	42*
	100 "	-	-	-	-	-	-	-	-	-	613*
2-AA	5 "	1422*	-	1264*	-	-	-	-	-	139	-
	20 "	-	-	-	-	768*	-	-	-	-	-
CPPM	250 "	-	-	-	-	-	-	510*	-	-	-

* Significantly different from the solvent control: greater than 2-fold increase over the solvent control; DNRB-HCl = Daunorubicin-HCl; 4-NTQLO = 4-Nitroquinoline-oxide; MTMC-C = Mitomycin-C; ANAD-HCl = Aminoacridine Hydrochloride; 2-AA = 2-Aminoanthracene; CPPM = Cyclophosphamide.

Results: continued

Treatment	Conc. (Per Plate)	Mean Number of His+ Revertant Colonies Per Plate									
		TA98		TA100		TA102		TA1535		TA1537	
		+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9	+S9	-S9
2nd Trial:											
Solvent Control		49	29	117	178	285	336	18	15	17	8
CGA 154281	20 ug	44	32	97	161	290	309	16	12	13	5
	78 "	55	35	120	156	285	311	15	13	14	5
	313 "	64	31	102	135	280	276	13	13	13	10
	1250 "	52	28	122	157	184	191	12	13	11	11
	5000 "	97*	5	135	186	165	136	12	13	4	12
Positive Controls:											
DNRB-HCl	5 ug	-	701*	-	-	-	-	-	-	-	-
	10 "	-	1172*	-	-	-	-	-	-	-	-
4-NTQLO	0.125 "	-	-	-	708*	-	-	-	-	-	-
	0.25 "	-	-	-	1189*	-	-	-	-	-	-
MTMC-C	0.5 "	-	-	-	-	-	1139*	-	-	-	-
	1.0 "	-	-	-	-	-	1320*	-	-	-	-
Sodium Azide	2.5 "	-	-	-	-	-	-	-	785*	-	-
	1.0 "	-	-	-	-	-	-	-	981*	-	-
ANAD-HCl	50 "	-	-	-	-	-	-	-	-	-	168*
	100 "	-	-	-	-	-	-	-	-	-	1150*
2-AA	5 "	1493*	-	792*	-	-	-	-	-	71*	-
	20 "	-	-	-	-	625*	-	-	-	-	-
CPPM	250 "	-	-	-	-	-	-	348*	-	-	-
3rd Trial:											
Solvent Control		59	35	-	-	-	-	-	-	-	-
CGA 154281	500 ug	60	32	-	-	-	-	-	-	-	-
	1000 "	65	47	-	-	-	-	-	-	-	-
	2000 "	87	74*	-	-	-	-	-	-	-	-
	4000 "	98*	83*	-	-	-	-	-	-	-	-
	8000 "	59	102*	-	-	-	-	-	-	-	-
Positive Control:											
DNRB-HCl	5 "	63	707*	-	-	-	-	-	-	-	-
	10 "	1511*	1165*	-	-	-	-	-	-	-	-

* Significantly different from the solvent control: greater than 2-fold increase over the solvent control.

Findings:

1. Based on the results obtained from the preliminary toxicity test, slight reduction in the growth of background bacteria was observed at 5000 ug/plate. Therefore, the concentration of 5000 ug/plate was selected as the highest dose for this study (Trials 1 and 2).

Findings: continued

2. The spontaneous revertant colonies for each of these five strains of Salmonella typhimurium (TA98, TA100, TA102, TA1535, and TA1537) were found within the normal range of His^+ revertant colonies recommended by the Ames test (1975).

3. The strain specifc control compounds (Daunorubicin-HCl, 4-Nitroquinoline-oxide, Mitomycin-C, Sodium Azide and Aminoacridine Hydrochloride) and the positive control compounds to ensure the efficacy of the activation (2-Aminoanthracene and Cyclophosphamide) in this study have given the positive responses as expected.

4. In the experiments performed without microsomal activation on strain TA98, significant increases in the number of revertant colonies were observed in the third trial at the concentrations of 2000, 4000, and 8000 ug/plate (dose-related increases). In the experiments carried out with microsomal activation, a weak increase in the number of revertant colonies was also noted in the 2nd and 3rd trials at the concentrations of 4000 and 5000 ug/plate. No evidence of mutagenic potential was observed in other tester strains in this study.

Evaluation:

Under the test conditions reported, the test compound, CGA 154281 Technical, is considered mutagenic in the Ames Salmonella/Mammalian Microsomal Mutagenicity Test at the concentrations tested. However, a minor deficiency with respect to the density of grown cultures (i.e., 1-2 x 10^9 cells per ml) in reporting of this study was noted. This study is considered acceptable.